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STANDARDIZATION OF PHARMACEUTICAL PREPARATIONS; THE JATYADI TAILA & JATYADI GHRITA AND THEIR ANTIMICROBIAL STUDY

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ABSTRACT: Ghrita and Taila kalpana are important preparations which use the process of incorporating drugs in oily particle to target them to their site of action. The Jatyadi Taila and Jatyadi Ghrita are important preparations which are used very commonly in Ayurvedic field of surgery as Vrana Ropak (wound healer).

This study explores the rationality behind the Paka Kala, Murcchana and its utility with chemical analysis and comparative antimicrobial study of Jatyadi Taila & Jatyadi Ghrita.

Jatyadi Taila and Jatyadi Ghrita has an effective antimicrobial activity against Staphylococcus and Pseudomonas in both Diffusion methods (Kirby- Bauer disc diffusion method and Stokes disc diffusion method) and Dilution methods (Broth dilution method & Agar dilution method). The Acid value, Saponification value, Peroxide value, Ester value, thin Layer Chromatography of two samples was evaluated.

INTRODUCTION

The Jatyadi Taila and Jatyadi Ghrita have been used in Ayurvedic System of surgery from older times. It is famous mainly for its anti-inflammatory, analgesic effects & healing property for which, these are used externally in Vrana treament. It is also mentioned that it is good for Nadi Vrana, Marmaashrita Vrana, Deep seated Vrana & Dusta Vrana (infected wound). An Antimicrobial is an agent that kills microorganism or inhibits their

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growth and Antibiotics are useful for the treatment of infectious disease in situations where the normal host defense cannot destroy pathogens. Antimicrobial agents differ not only in there action and activity but also in their distribution, metabolization and excretion from the body. When immediate antimicrobial therapy is essential there is no time to culture and identify the disease causing agent. So drug (Jatyadi Taila and Jatyadi Ghrita) specificity was checked with methods by Kirby- Bauer disc diffusion method, Stokes disc diffusion method, Broth dilution method and Agar dilution method.

AIM AND OBJECTIVE

The main aim of this study to Chemical Analysis of Jatyadi Taila & Jatyadi Ghrita and Comparative antimicrobial study of Jatyadi Taila & Jatyadi Ghrita.

MATERIAL AND METHODS

Methods were used for Tila Taila Murcchana and Ghrita Murcchana according to Bhaishjya Ratnavali Jvaradhikar, and Preparation of Jatyadi Taila and Jatyadi Ghrita according Sharangdhar samhita in present study.

After preparation of Jatyadi Taila and Jatyadi Ghrita the samples were taken for chemical analysis, Sample (A) - Jatyadi Taila and Sample (B) - Jatyadi Ghrita. Samples were carried out at Oasis Test House Limited, Jaipur. (No.-M1897/2010 and M1898/2010).

Comparative antimicrobial study of Jatyadi Taila & Jatyadi Ghrita done through antimicrobial sensitivity tests were takes place by two types of methods as below:

· **Diffusion methods**

A. Kirby- Bauer disc diffusion method

B. Stokes disc diffusion method

· **Dilution methods**

A. Broth dilution method

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B. Agar dilution method

Table-1 Maintenance of culture :

The stock cultures were maintained on the following media;-

S.N.	**Bacterial Species**	**Type of Bacterial media used**
1.	Staphylococcus aureus	5% blood agar, Nutrient Agar
2.	Pseudomonas aeruginosa	Nutrient Agar, Mac Conkey Agar

Strains of Bacterial species were selected by their various antibiograms prevalent in local population.

First method-

Step I- First of all organism were selected from sample that was from an infected wound. It was inoculated on suitable media & incubated at 35^0- 37^0C for 18-24 hrs. We perform coagulase test and organism was positive as the colour of medium is changed pink to yellow.

Step II- A single colony of Staphylococcus aureus and Pseudomonas aeruiginosa was inoculated on glucose broth separately and incubated for 4 hrs at 35^0- 37^0C.

Step III- Now a well was created of 6 mm diameter with cup-well technique on plate then it was filled with each sample of Jatyadi Taila & Jatyadi Ghrita & other antibiotics discs are also placed on the plate for comparative study. After 24 hours of incubation at 35^0- 37^0C, the diameter of zone of complete growth inhibition (in mm) was measured as per standard procedure described vide information.

OBSERVATIONS

:

(A) Chemical analysis

Zone of inhibition (ZOI) of both sample drugs is as follows.

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Staphylococcus aureus

Table no.

Bacterial Strains	ZOI	
	A	B
1	2 mm	3 mm
2	3 mm	2 mm
3	1 mm	1 mm
4	2 mm	2 mm

Pseudomonas aeruginosa

Table no.

Bacterial Strains	ZOI	
	A	B
1	13 mm	8 mm
2	12 mm	6 mm
3	14 mm	7 mm
4	13 mm	8 mm

In case of Staphylococcus aureus the result was unsatisfactory as the ZOI were very small means intermediate sensitive in some strains & in some case it was resistant.

In case of Pseudomonas aeruiginosa the result was partial positive because ZOI was not very clear in compare to other antibiotics. But it was better with Jatyadi Taila in comparison to Jatyadi Ghrita.

Second Method-

Step I

Firstly organism was selected from sample that was from an infected wound. It was inoculated on suitable media & incubated at 35^0- 37^0C for 18-24 hrs. We perform coagulase test and organism was positive as the colour of medium is changed pink to yellow.

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Step II

A single colony of Staphylococcus aureus and Pseudomonas aeruiginosa was inoculated on glucose broth separately and incubated for 4 hrs at 35^0- 37^0C.

Step III

Drug dilution :

Initially 10 gram of each sample was weighed separately and diluted in 20 ml Dimethyl Pharmamide. It means the concentration of drug was 500 mg /ml.

Preparation of different concentration:

Both diluted sample solution was measured by pipette and placed in sterile test tube containing measured glucose broth solution. Both samples were taken in separate test tube according to following table.

Jatyadi Taila sample 'A'

Test tubes containing sample of 'A'					
Drug	0.5 ml	1 ml	2 ml	3 ml	4 ml
Medium	9.5 ml	9 ml	8 ml	7 ml	6 ml
Conc.	250 mg/ml	500 mg/ml	1000mg/ml	1500mg/ml	2000mg/ml

Jatyadi Ghrita sample 'B'

Test tubes containing sample of 'B'					
Drug	0.5 ml	1 ml	2 ml	3 ml	4 ml
Medium	9.5 ml	9 ml	8 ml	7 ml	6 ml
Conc.	250 mg/ml	500 mg/ml	1000mg/ml	1500mg/ml	2000mg/ml

Now a small loop full inoculum (4 hrs cultured) were mixed in each test tube respectively. Then each test tube was shook well and incubated at 35^0- 37^0C for 18-24 hrs.

Next day turbidity was visible in above dilutions. Now a loop full sample were inoculated on nutrient agar of all respective dilution separately and incubated at 35^0- 37^0C for 18-24 hrs.

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For each bacteria and drug sample, different plates were taken and then observation was taken with precautions.

Next day observation was noted according to following criteria:

S.n.	Sensitivity	Drug concentration	Bacterial growth
1.	0	250 mg/ml	++++
2.	Mild	500 mg/ml	+++
3.	Moderate	1000 mg/ml	++
4.	Good	1500 mg/ml	+
5.	Best	2000 mg/ml	0

(B) Comparative antimicrobial study

Scoring method is followed to simplify the observations in Comparative antimicrobial study.

Sample 'A' and 'B'

S.n.	Sensitivity	Drug concentration	Bacterial growth	Score
1.	0	250 mg/ml	++++	1
2.	Mild	500 mg/ml	+++	2
3.	Moderate	1000 mg/ml	++	3
4.	Good	1500 mg/ml	+	4
5.	Best	2000 mg/ml	0	5

In case of Staphylococcus aureus the result was poor at concentration 250 mg/ml but it was best at 2000 mg/ml.

In case of Pseudomonas aeruginosa the result was poor at concentration 250 mg/ml but it was best at 2000 mg/ml.

The comparative observation can be represented as follows:

Table no.

Drug	Jatyadi Taila	Jatyadi Ghrita

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		Score		Score	
s.n.	**concentration**	**Staph.**	**Pseudo.**	**Staph.**	**Pseudo.**
1.	250 mg/ml	1	2	1	2
2.	500 mg/ml	1	4	2	5
3.	1000 mg/ml	4	5	3	5
4.	1500 mg/ml	4	5	4	5
5.	2000 mg/ml	5	5	5	5

The above result can be summarized as follows:

Viewing the result we can say that Staphylococcus aureus showed greater antibacterial activity towards Jatyadi Taila than Jatyadi Ghrita but in case of Pseudomonas aeruginosa, Jatyadi Ghrita showed greater antibacterial activity than Jataydi Taila.

Jatyadi Taila-

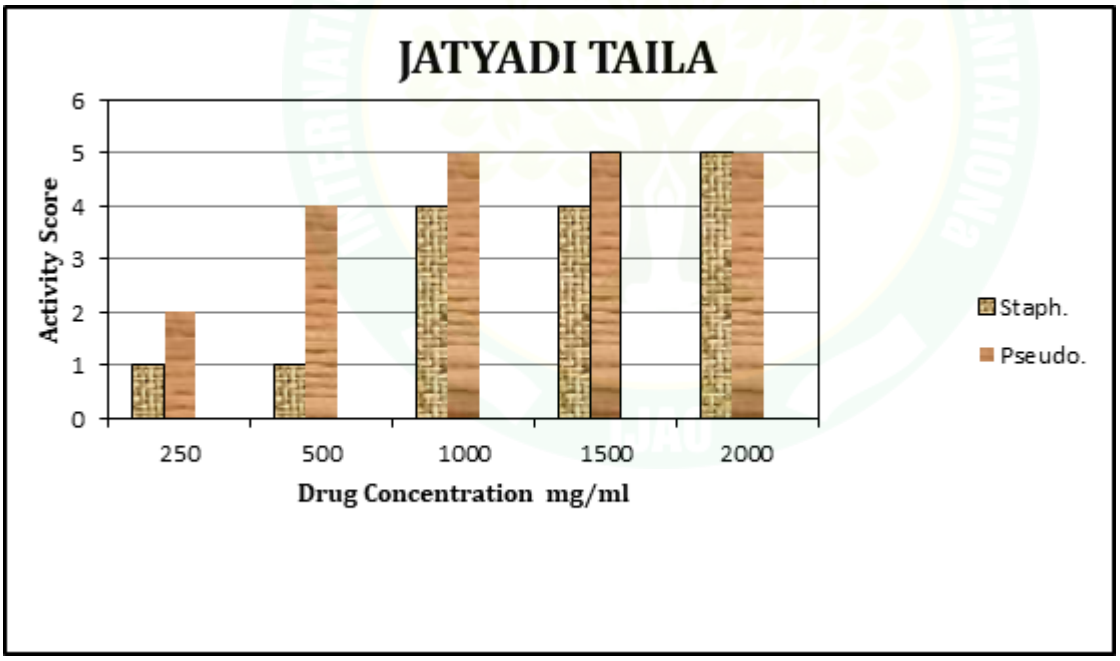


Figure 1: figure 1

Jatyadi Ghrita-

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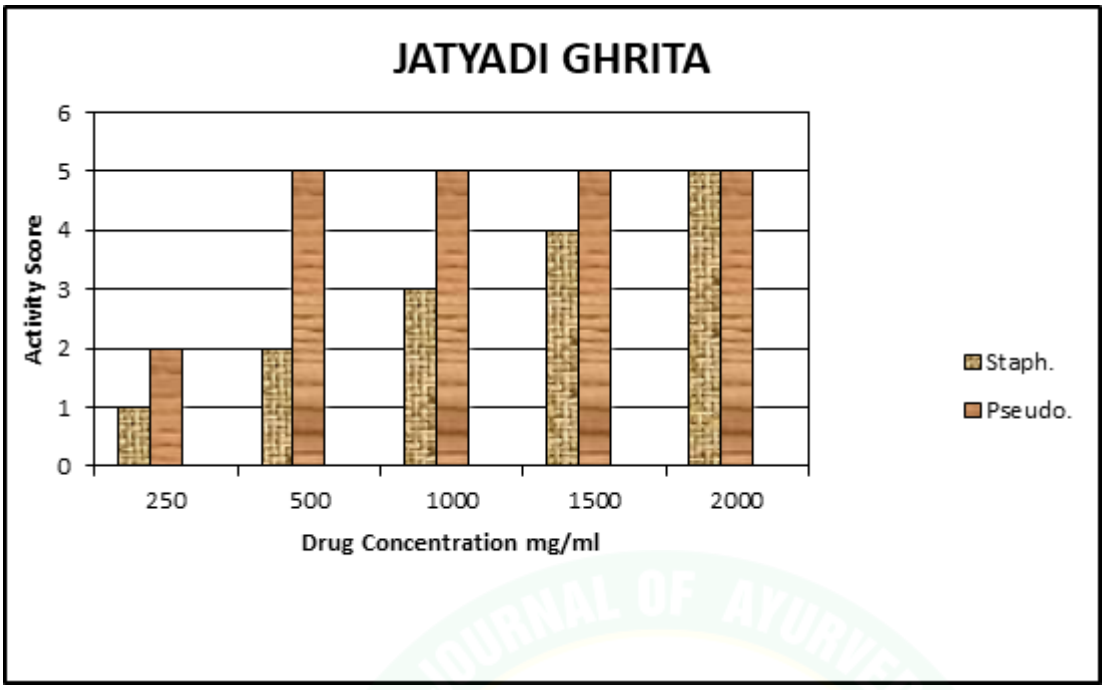


Figure 2: figure 2

RESULTS

The results of standardization of preparations were show that the Jatyadi taila had Iodine value 97.17, peroxide value 5.07, saponification value 186.78, acid value 2.20, ester value 184.57, Rf value 0.92 and in the Jatyadi Ghrita had Iodine value 64.84, peroxide value 13.51, saponification value 124.69, acid value 2.89, ester value 121.79, Rf value 0.91.

Table no. - Chemical Analysis of Jatyadi Taila & Jatyadi Ghrita

S.N.	Parameter	Observation	
		Jatyadi Taila	Jatyadi Ghrita
1.	Appearance	Reddish Brown Coloured viscous oil	Greenish Coloured semi solid mass
2.	Odour	Characteristic	Characteristic
3.	Touch	Oily	Greasy
4.	Iodine Value	97.17	64.84
5.	Peroxide value	5.07	13.51

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6.	Saponification value	186.78	124.69
7.	Acid value	2.20	2.89
8.	Ester value	184.57	121.79
9.	Moisture	0.079%	0.023%
10.	Thin Layer Chromatography-Rf Value	0.92	0.91

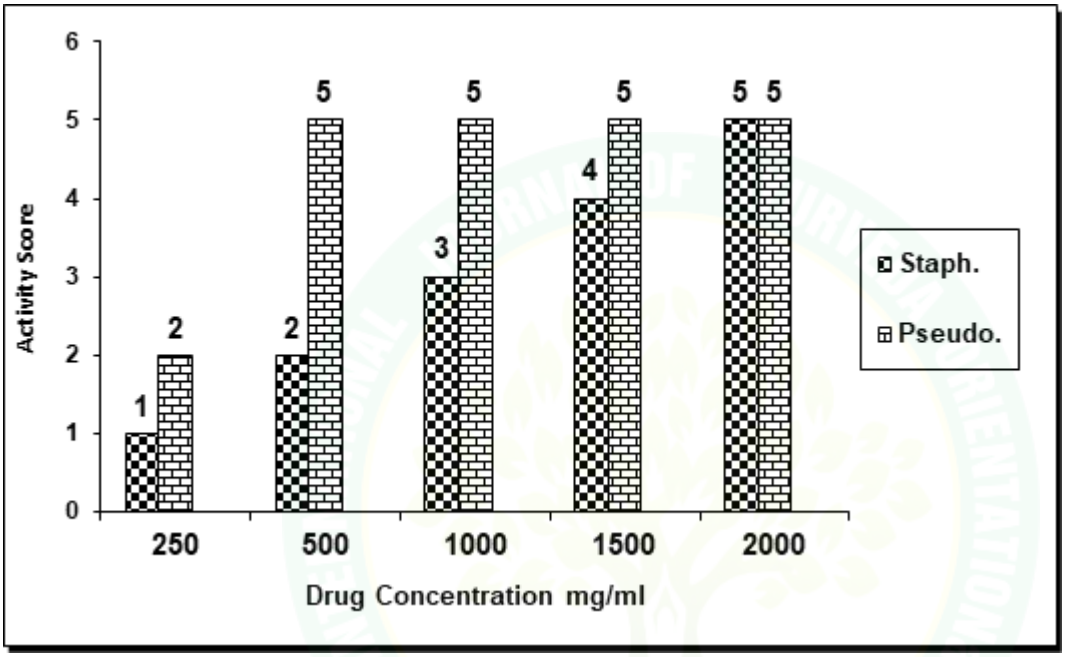
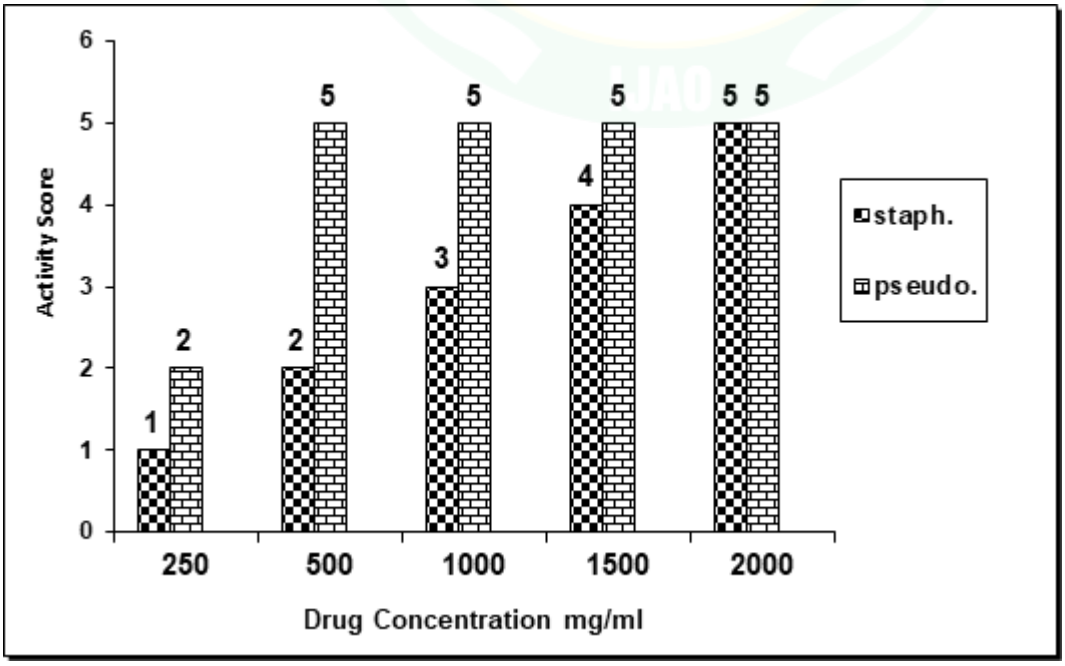


Figure 3: figure 3



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Figure 4: figure 4

CONCLUSION

After analysing the observations Jatyadi Ghrita showed greater antimicrobial activity than Jatyadi Taila on Pseudomonas but Jatyadi Taila showed greater antimicrobial activity than Jatyadi Ghrita on Staphylococcus.

The study show that Jatyadi Ghrita is better than Jatyadi Taila in the case of Pseudomonas infected wound and Jatyadi Taila is better than Jatyadi Ghrita in Staphylococcus infected wound.

A combination regime may be more effective in infected wound & in resistant cases than the drugs used alone, but such definitive statements would require more detailed studies to be carried out on such regimes to exclude any long term adverse effects as experienced by the patients or on the evidence of biochemical tests.

Present study may prove & be helpful to develop a good antimicrobial liniment. Thus the present work needs further studies not only in-vitro but also in-vivo on larger groups of the patients to reach more definitive conclusions.

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